# A method to choose water depths for zooplankton samples in lakes 

Brian Rippey © ${ }^{(1 *}{ }^{1 *}$ Katrina A. Macintosh ${ }^{\bullet},^{1,3}$ Yvonne McElarney ${ }^{(\odot)}{ }^{2}$ Richard Douglas ${ }^{1}$<br>${ }^{1}$ School of Geography and Environmental Sciences, Ulster University, Coleraine, UK<br>${ }^{2}$ Agri-Food and Biosciences Institute, Belfast, UK<br>${ }^{3}$ DairyNZ, Lincoln, New Zealand


#### Abstract

As methods in the literature to sample zooplankton in lakes mostly offered general guidance on the sample depths, a new one was developed. Using the principle of volume-weighted sampling of the lake volume and an empirical function for the hypsometric curve, formulae for the volumes and areas of five equal sections of the lake were derived, which were then used to calculate section mean depths. Vertical net hauls taken at the mean depths are combined using a relation between their mean depths to produce one unbiased composite sample of the zooplankton. While generic formulae were derived, starting values for the depths that divide the lake volume into five equal sections are needed in order to apply the method, which then optimizes the depths; the method is implemented in a spreadsheet. The method was applied to four hypothetical lakes of maximum depth 12 m that cover a wide variation of lake form and how the sample depths vary with form was described; as lake form becomes more convex, the sample depths decrease, reflecting that more of the lake volume is at shallower depth. The method was used to estimate the whole-lake abundance of zooplankton in 51 lakes and no practical difficulties were encountered. It can be used in lakes up to a few tens of $\mathrm{km}^{2}$ in area.


Describing the key characteristics of zooplankton is commonly needed when investigating the behavior of lakes, as this ecological group is important in the lake food web. Examples where zooplankton were involved are: reduction of the chlorophyll (Chl) a concentration through grazing of phytoplankton, described using zooplankton biomass (Hanson and Peters 1984; Kamarainen et al. 2008) or the presence of large bodied Daphnia (Mazumder 1994); reduction in the mean body size of all zooplankton and of cladocerans (Mills and Schiavone Jr (1982) or of Daphnia (Hessen et al. 1995) used to indicate predation by fish; and the key role of zooplankton in the biomanipulation of lakes through top down control (Brett and Goldman 1996; Jeppesen et al. 2000).

We wished to characterize the zooplankton as a part of a synoptic description of 51 Irish lakes (Table 1), as they are an important component of the lake food web (Brett and

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Goldman 1996; Jeppesen et al. 2003). However, when reviewing the sampling methods in the literature, we found that they varied and most provided only general guidance. For example, Bottrell et al. (1976) give general advice that the sampling should suit the situation, with the decision on sites based on systematic sampling, and Galbraith Jr and Schneider (2000) advise four samples arranged systematically in four quadrants.

The recommendations of the US EPA (2013) are also general, with no mention of the number of sites or their location, other than two sampling tows per station. The European standard (EN 15110: 2006 2006) advises that the number and location of the sampling sites should be determined according to the aim of the study, with the deepest or central area suggested and that the sampling program should be adapted to the lake morphology and be stratified.

Blomqvist (2001) completed a comprehensive analysis of how the vertical and horizontal variability in lakes can be accounted for and proposed a sampling method to characterize chemical concentrations and plankton abundance. The basis of the method is volume-weighted sampling of horizontal layers of the water column so that no depth is over- or underrepresented. Based on a single lake basin, typically five sites, one from the center and one each from four quadrants, are used and the number of samples from each horizontal

Table 1. Summary of the size of 51 small Irish lakes, including the $q$ exponent in Eq. 1.

|  | Average | Median | $\mathbf{1 0}^{\text {th }}$ percentile | $\mathbf{9 0}^{\text {th }}$ percentile |
| :--- | :---: | :---: | :---: | :---: |
| Area, ha | 109 | 36.2 | 6.60 | 203 |
| Mean depth, $m$ | 4.1 | 4.0 | 1.4 | 7.2 |
| Maximum depth, $m$ | 13.0 | 12.0 | 5.2 | 24.2 |
| $q$, dimensionless | 2.41 | 1.99 | 1.00 | 4.42 |

layer is determined by the volume-depth relationship in the lake. The layer thickness varies from 0.5 to 4 m depending on the maximum water depth of the lake.

Girard et al. (2007) also provided a detailed description of a method; vertical net hauls from four to seven sites chosen "to minimize the deviation of the cumulative fraction of the total haul lengths from the fraction of the total lake volume that were between increments of depth. In other words, a composite sample was formed that corrected for the diminution of lake volume with depth." While the sampling depths used for each lake are given, how they were derived was not. Nevertheless, the basis of the method is clear; volume-weighted sampling of vertical slices of the lake water so that no depth is over- or underrepresented. The method of Rusak and Montz (2009) is also based on a similar principle, "hypsometrically pooled (combined in proportion to the volume that the particular depth stratum contributes to the total lake volume)," although, again, no details of how this was achieved were given.

Overall, the method described by Blomqvist (2001) and the basics of the methods of Girard et al. (2007) and Rusak and Montz (2009) are based on the same principle; sampling the water column at between four and seven sites so that the overall sample is weighted according to the volume-depth relationship in the lake. There are slight differences, as discrete water samples are taken from water layers in the methods of Blomqvist (2001) and Rusak and Montz (2009), while Girard et al. (2007) use vertical net hauls. Nevertheless, all methods sample the upper, middle, and lower water layers in the same proportion as their contribution to the total lake volume.

Blomqvist (2001), based on one lake of 75 ha area, Girard et al. (2007), based on 72 lakes with areas from 5 to 79 ha, and Rusak and Montz (2009), based on four lakes with areas from 37 to 1608 ha, suggest sampling the lake at between four and seven sites and this range is supported by others (Downing et al. 1987; Pace et al. 1991). Patalas and Salki (1993) investigated the influence of lake size on the spatial variation of zooplankton abundance and found three to six stations appropriate in small to medium sized ( $<60 \mathrm{~km}^{2}$ ) lakes. Galbraith Jr and Schneider (2000) suggest four samples for a single lake basin.

Based on this review, there is a need for a well-defined method to select the water depths to sample the zooplankton in lakes that can be easily applied using only basic information. If the lakes on which the review of the number of replicates are used, then the method should apply to lakes up to

1608 ha in area and possibly $60 \mathrm{~km}^{2}$ if they have a single basin. Spatial variability in zooplankton species density is likely to be greater in larger lakes (Patalas and Salki 1993) and in ones with multiple basins, so it seems best to restrict the use of the method presented to lakes up to a few tens of $\mathrm{km}^{2}$ in area if between four and seven replicate samples are used.

## Use materials and procedures

The method developed to characterize the zooplankton was based on the principle underlying the methods of Blomqvist (2001), Girard et al. (2007), and Rusak and Montz (2009); volume-weighted sampling of the lake so that no part is under- or overrepresented. We chose five samples within each lake, as this is the typical number suggested by others and composited vertical net hauls to characterize the zooplankton with one representative sample.

The work to develop the method consisted of three stages: describing the lake hypsometric curve, selecting water depths of the five samples, and compositing the net haul samples.

## Lake hypsometric curve

Blomqvist (2001) employed two relationships proposed by Hakanson (1981) for the variation with depth of the water layer volume that depend on whether the lake form (hypsometric curve) is linear to concave or convex, using a linear or parabolic approximation of the volume of the horizontal water layers, respectively.

We used the empirical function between lake area and water depth proposed by Imboden (1973) as it allows other lake form properties to be described and so made generalization of the method easier. It was used by Livingstone and Imboden (1996) to model deoxygenation in lake hypolimnia. It was the basis of a model of the vertical profile of radon- 222 used to estimate the vertical and horizontal eddy diffusion coefficients in a lake (Imboden and Joller 1984). A final example of its use was to correct the carbon accumulation rate measured in sediment at the deepest part of a lake in order to estimate the whole-basin rate (Ferland et al. 2014).

The hypsometric curve for a lake can be described empirically by the function (Livingstone and Imboden 1996):

$$
\begin{equation*}
A(Z)=A(0)\left(1-\left(Z / Z_{\mathrm{MAX}}\right)\right)^{q} \tag{1}
\end{equation*}
$$

where $A(Z)$ is the area at water depth $Z, A(0)$ is the surface area of the lake ( $Z=0$ ), $Z$ is the depth of water, positive downward


Fig. 1. The variation of area $(A(Z))$ at water depth $Z$ for six values of the nondimensional exponent q (Eq. 1). The area is expressed as a proportion of the lake surface area $(A(0))$ and water depth given as a proportion of the maximum depth $\left(Z_{\text {MAX }}\right)$.
from lake surface, $Z_{\text {max }}$ is the maximum depth of water, and $q$ is a nondimensional exponent.

The general behavior of the lake form function is displayed in Fig. 1. The exponent $q$ is derived by fitting the function to the observed variation of $A(Z)$ with $Z$, the hypsometric curve of the lake. As with any model, it should capture the general pattern of how area varies with depth but may only be an approximate characterization in some lakes; it could also be more accurate at some depths and less so at others and additional considerations may be needed in lakes with multiple basins. Examples of how good a fit the function is in small lakes are provided in Supplemental Information.

If a bathymetric map is not available, an approximation of the function can be produced, as shown next (Eq. 5), by using estimates of the mean and maximum depths, based on spot depth observations; the more spot depths available, the better the approximation should be.

First, Eq. 1 is used to derive the formula for the variation with depth ( $Z$ ) of the volume from depth $Z$ to $Z_{\text {max }}(V(Z))$ (Eq. 2), as shown in the Supplemental Information.


Fig. 2. The frequency distribution of $q$ value for a global lake data set, $n=905$.

$$
\begin{equation*}
V(Z)=\frac{Z_{\mathrm{MAX}} A(0)}{(q+1)}\left(1-\frac{Z}{Z_{\mathrm{MAX}}}\right)^{q+1} \tag{2}
\end{equation*}
$$

Using Eq. 2, the total volume of the lake $(V(Z)$ at $Z_{\text {MAX }}$ is:

$$
\begin{equation*}
\text { Volume of lake }(V(0))=\frac{Z_{\operatorname{MAX}} A(0)}{(q+1)} \tag{3}
\end{equation*}
$$

As the lake mean depth ( $Z_{\text {MEAN }}$ ) is lake volume (Eq. 3) divided by surface area $(A(0)$ ), the mean depth is

$$
\begin{equation*}
Z_{\mathrm{MEAN}}=\frac{Z_{\mathrm{MAX}}}{(q+1)} \tag{4}
\end{equation*}
$$

Rearranging allows $q$ to be calculated from estimates of the mean and maximum depths;

$$
\begin{equation*}
q=\left(Z_{\mathrm{MAX}} / Z_{\mathrm{MEAN}}\right)-1 \tag{5}
\end{equation*}
$$

Applying Eq. 5 to a database of world lakes (ILEC World Lake Database; https://wldb.ilec.or.jp) gives the following statistics for $q\left(n=905\right.$ lakes; Fig. 2): minimum $0.09 ; 10^{\text {th }}$ percentile 0.64 ; mean 1.74 ; median $1.50 ; 90^{\text {th }}$ percentile 3.16 ; maximum 10.50 .
$q$ values less than 1.0 are found in concave form lakes and greater than 1.0 in convex (Fig. 1). As the majority of these q values are greater than 1.0 ( $72.7 \%$ ), most of the lakes have a convex internal form, which was also found by Hakanson (1977). A summary of the variation of $q$ in the 51 Irish lakes is given in Table 1.

## Selecting water depths of the five samples

The principle of selecting the samples is to collect five so that no part of the lake water is under- or overrepresented.


Fig. 3. Variation of lake area with depth for a lake of maximum depth 12 m and $q$ value 2.0. Full vertical lines indicate the five sections that divide the lake into five equal volumes at water depth $\mathrm{Z} 1, \mathrm{Z} 2, \mathrm{Z3}$, and Z 4 (indicated by horizontal dotted lines). The vertical dashed lines show the mean depths of the five sections.

This can be achieved by dividing the lake hypsometric curve into five vertical sections of equal volume and collecting a sample at the mean depth of each section; the sections could be considered slices of the hypsometric curve (Fig. 3) or rough concentric "doughnuts" looking vertically on to the lake surface. A vertical net haul taken at the average depth of each section filters zooplankton from above the sediment surface to the water surface and the five samples are then composited.

The depths that divide the lake volume into the five equal sections, Z1, Z2, Z3, and Z4, are illustrated in Fig. 3 and listed in Table 2 along with the nomenclature used for the area, volume and mean depth of the five sections.

As the volumes of the sections are to be equal, then,

$$
\begin{equation*}
\mathrm{VZ} 1=\mathrm{VZ} 2=\mathrm{VZ} 3=\mathrm{VZ} 4=0.2 V(0) \tag{6}
\end{equation*}
$$

From the volume and area of each section, the mean depth can be calculated (Eq. 7) and this is the sample depth for the vertical net haul.

$$
\begin{equation*}
Z_{\mathrm{MEAN}}=\mathrm{VZ} / \mathrm{AZ} \tag{7}
\end{equation*}
$$

where VZ and AZ are the volume and area of each section.

The areas of the sections are derived using Eq. 1 to calculate the area at each of the depths that divide the sections and subtracting the areas of the deeper sections as follows (Table 2).

AZ1 is calculated using Eq. 1 with $Z=Z 1$; AZ2 as (Eq. 1 with $\mathrm{Z}=\mathrm{Z} 2$ ) - AZ1; AZ3 as (Eq. 1 with $Z=\mathrm{Z}$ ) - AZ1 - AZ2; AZ4 as (Eq. 1 with $Z=\mathrm{Z} 4$ ) - AZ1 - AZ2 - AZ3 and; AZ5 as $V$ (0) - AZ1 - AZ2 - AZ3 - AZ4.

The volumes of the sections need to be derived in two stages. First, calculate the cumulative volumes of the sections starting with Section 1 (Table 2). VZ1 is the volume below depth Z1 plus the volume that sits on top of the lake area at water depth Z 1 . The volume below Z 1 is calculated using Eq. 3 with $Z=\mathrm{Z} 1$ and the volume that sits on top is the product of Z 1 and the area at water depth Z1 (Eq. 1).

The volume of Sections 1 and 2 is the volume below depth Z2 plus the volume that sits on top of the lake area at water depth Z2. The volume below Z 2 is calculated using Eq. 3 with $Z=\mathrm{Z} 2$ and the volume that sits on top is the product of Z 2 and the area at water depth Z2 (Eq. 1).

The volume of Sections 1-3 is the volume below depth Z3 plus the volume that sits on top of the lake area at water depth Z3; the volume of Sections $1-4$ is the volume below depth $Z 4$ plus the volume that sits on top of the lake area at water depth Z4.

Second, using these cumulative volumes, the volume of each section is as follows:

VZ2 is the volume of Sections 1 and 2 minus VZ1.
VZ3 is volume of Sections 1-3 minus VZ1 minus VZ2.
VZ4 is volume of Sections 1-4 minus VZ1 minus VZ2 minus VZ3.
VZ5 $=V(0)-\mathrm{VZ1}-\mathrm{VZ2}-\mathrm{VZ3}-\mathrm{VZ} 4$.

The mean depth of the five sections can now be calculated using their area and volume (Eq. 7) and these are the water depths for the vertical net haul samples (Table 2). The length of the hauls would be a little less so as not to disturb the sediment.

While application of the principle gives the water depths of the samples, they cannot be derived without an initial estimate of the values of $\mathrm{Z} 1, \mathrm{Z} 2, \mathrm{Z} 3$, and Z 4 . A formal solution to deriving the sample depths seems impossible, given the use of five vertical section, the areas and volumes of which are derived by subtraction of cumulative areas and volumes to calculate their mean depths; it is the use of five vertical sections of the lake that creates the difficulty of a formal solution. We, therefore, implemented the method in the spreadsheet, a method to choose water depths for zooplankton samples in lakes (Supplemental Information). Based on initial values for $\mathrm{Z} 1, \mathrm{Z} 2, \mathrm{Z} 3$, and Z 4 , in stages the nonlinear regression add-in program Solver in Microsoft Excel (Brown 2001) finds new values that satisfies the criterion that the volume of each section is $0.2 V(0)$ (Eq. 6). The initial estimates are $\mathrm{Z} 1=0.8 Z_{\mathrm{MAX}}, \mathrm{Z} 2=0.6 Z_{\mathrm{MAX}}, \mathrm{Z} 3=0.4 Z_{\mathrm{MAX}}$, and $\mathrm{Z} 4=0.2 Z_{\mathrm{MAX}}$.

Table 2. Nomenclature used for the depths dividing the five vertical lake sections and the area, volume, and mean depth of the sections. The formulae used to calculate the volume, area, and mean depths of the sections are also shown.

| Section | 1 | 2 | 3 |  | 4 |  | 5 |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Depth dividing sections |  |  |  | Z3 |  | Z4 |  |
| Area of section | AZ1 | AZ2 | AZ3 |  | AZ4 |  | AZ5 |
| Volume of section | VZ1 | VZ2 | VZ3 |  | VZ4 |  | VZ5 |
| Mean depth of section | D1 | D2 | D3 |  | D4 |  | D5 |
| AZ1 | Eq. 1 with $Z=Z 1$ |  |  |  |  |  |  |
| AZ2 | (Eq. 1 with $Z=$ Z2) - AZ1 |  |  |  |  |  |  |
| AZ3 | (Eq. 1 with $Z=Z 3$ ) - AZ1 - AZ2 |  |  |  |  |  |  |
| AZ4 | (Eq. 1 with $Z=\mathrm{Z} 4$ ) - AZ1 - AZ2 - AZ3 |  |  |  |  |  |  |
| AZ5 | A(0) - AZ1 - AZ2 - AZ3 - AZ4 |  |  |  |  |  |  |
| VZ1 | (Eq. 3 with $Z=Z 1)+(Z 1 \times$ Area at $Z=Z 1)$ |  |  |  |  |  |  |
| VZ2 | (Eq. 3 with $Z=Z 2)+(Z 2 \times$ Area at $Z=Z 2)-V Z 1$ |  |  |  |  |  |  |
| VZ3 | (Eq. 3 with $Z=\mathrm{Z} 3)+(\mathrm{Z3} \times$ Area at $\mathrm{Z}=\mathrm{Z} 3)-\mathrm{VZ1}-\mathrm{VZ2}$ |  |  |  |  |  |  |
| VZ4 | (Eq. 3 with $Z=Z 4)+(\mathrm{Z} 3 \times$ Area at $\mathrm{Z}=\mathrm{Z} 4)-\mathrm{VZ1}-\mathrm{VZ2}-\mathrm{VZ3}$ |  |  |  |  |  |  |
| VZ5 | $V(0)-\mathrm{VZ1}-\mathrm{V} Z 2-\mathrm{V} Z 3-\mathrm{V} 24$ |  |  |  |  |  |  |
| D1 | VZ1/AZ1 |  |  |  |  |  |  |
| D2 | VZ2/AZ2 |  |  |  |  |  |  |
| D3 | VZ3/AZ3 |  |  |  |  |  |  |
| D4 | VZ4/AZ4 |  |  |  |  |  |  |
| D5 | VZ5/AZ5 |  |  |  |  |  |  |

Table 3. Variation of water depth ( $m$ ) of the five vertical net hauls for four hypothetical lakes with varying mean depth ( $Z_{\text {MEAN }}$ ) and $q$ value. The lake areas are 10 ha and maximum depth 12 m .

| Lake | $\boldsymbol{Z}_{\text {MEAN }} \mathbf{m}$ | $\boldsymbol{q}$ | Haul $\mathbf{1}, \mathbf{m}$ | Haul $\mathbf{2 ,} \mathbf{m}$ | Haul 3, $\mathbf{m}$ | Haul 4, $\mathbf{m}$ | Haul 5, $\mathbf{m}$ |
| :--- | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 1 | 6.5 | 0.8 | 11.6 | 10.5 | 9.0 | 7.0 | 3.0 |
| 2 | 4.5 | 1.7 | 10.3 | 8.3 | 6.6 | 4.8 | 1.9 |
| 3 | 3.5 | 2.4 | 9.0 | 6.9 | 5.3 | 3.7 | 1.4 |
| 4 | 2.5 | 3.8 | 7.3 | 5.2 | 3.9 | 2.7 | 1.0 |

## Compositing the net haul samples

Vertical net hauls were deemed an appropriate sampling method (de Bernardi 1984; Mack et al. 2012) for characterizing the zooplankton as a component of the lake food web, given the focus of the investigation was the large herbivores (large cladocerans, mostly daphnids; Mazumder 1994) that graze on phytoplankton. The hauls were taken using a 30 cm diameter, 55 cm long conical plankton net of $250 \mu \mathrm{~m}$ mesh (EE and GB Nets, now part of NHBS), and 100\% filtration efficiency was assumed (Mack et al. 2012). Sampling was completed between 10:00 and 16:00 h.

Samples were taken in a random pattern at the selected depths. The first haul was taken at the appropriate depth closest to the deepest point of the lake, and then a random number between 0 and 360 used to choose bearings to the other four hauls and samples taken at the appropriate water depths.

The samples were preserved in $70 \%$ industrial methylated spirit. They could not be simply composited, as the lengths of
the hauls are different and so too the volumes of water filtered; the longer hauls, that is, Haul 1 and 2, would make up a disproportionally large contribution to the composite sample taxa densities, as they filter much larger volumes than the shorter hauls, that is, Haul 4 and 5. However, as the relationships between the five volumes filtered are identical to those between the length of the hauls, the proportions of the samples that need to be amalgamated are (haul length of the shallowest section (Haul 5)/(haul length); these are calculated in the spreadsheet, a method to choose water depths for zooplankton samples in lakes (Supplemental Information).

As the volumes of the net haul samples flushed from the net filter in the field varied, the samples were adjusted to 50 mL volume as follows. The zooplankton in each of the five samples were collected by filtering and transferred to a 50 mL tube and made up to 50 mL in $70 \%$ industrial methylated spirit. The proportions of the five net haul samples were prepared using the swirling flask-Stempel pipette method


Fig. 4. Variation of water depth of the five vertical net hauls for four hypothetical lakes with varying $q$ value. The lake areas are 10 ha and maximum depth 12 m . A lake with mean depth of 6.5 m has a $q$ value of $0.8,4.5 \mathrm{~m}$ has $1.7,3.5 \mathrm{~m}$ has 2.4 , and 2.5 m has 3.8 .

Table 4. The variation with mean depth ( $Z_{\text {MEAN }}$ ) and $q$ value of the proportions of the five haul samples needed to produce one composite sample for four hypothetical lakes of area 10 ha and maximum depth 12 m . The overall mean values and coefficient of variation (CV) are also given.

| Lake | $\boldsymbol{Z}_{\text {MEAN }}, \mathbf{m}$ | $\boldsymbol{q}$ | Haul $\mathbf{1 , ~} \mathbf{m}$ | Haul 2, $\mathbf{m}$ | Haul 3, $\mathbf{m}$ | Haul 4, $\mathbf{m}$ | Haul 5, $\mathbf{m}$ |
| :--- | :--- | :---: | :---: | :---: | :---: | :---: | :---: |
| 1 | 6.5 | 0.8 | 0.26 | 0.29 | 0.33 | 0.43 | 1.00 |
| 2 | 4.5 | 1.7 | 0.18 | 0.23 | 0.28 | 0.39 | 1.00 |
| 3 | 3.5 | 2.4 | 0.15 | 0.20 | 0.26 | 0.37 | 1.00 |
| 4 | 2.5 | 3.8 | 0.13 | 0.18 | 0.25 | 0.36 | 1.00 |
|  | Mean |  | 0.18 | 0.23 | 0.28 | 1.00 |  |
|  | CV, \% |  |  | 21.3 | 12.7 | 8.0 |  |

Table 5. The density (no. $\mathrm{L}^{-1}$ ) of zooplankton taxa in five vertical net hauls in a random pattern from across Legane Lough on 27 June 2012. The haul length, arithmetic mean density and CV and "composite" sample density calculated by weighting the relative volumes of water filtered. The lake area is 6.0 ha , maximum depth 12.8 m , mean depth 7.7 m , and $q$ value 0.7 .

| Haul | Length, $\mathbf{m}$ | Chaoborus spp. | Cyclops spp. | Daphnia longispina | Eudiaptomus gracilis |
| :--- | :---: | :---: | :---: | :---: | :---: |
| 1 | 12.6 | 0.2 | 0 | 13.6 | 10.2 |
| 2 | 11.8 | 0.2 | 0 | 23.3 | 9.7 |
| 3 | 10.4 | 0 | 0 | 23.3 | 12.7 |
| 4 | 8.4 | 0 | 0 | 13.1 | 17.8 |
| 5 | 3.7 | 0 | 0.2 | 18.38 | 13.1 |
| Arithmetic mean |  | 0.08 | 26 | 12.70 |  |
| CV, \% | 137 |  | 8.27 | 25 |  |
| "Composite" | 0.02 | 0.05 |  | 6.32 |  |

(McCauley 1984) and combined to produce one composite sample.

A subsample of between 1 and 5 mL of the composite sample, depending on the zooplankton density, was enumerated using an Olympus SX15 binocular microscope
and all animals counted. They were identified to the lowest taxonomic level using the Freshwater Biological Association keys to the British freshwater Cladocera (Scourfield and Harding 1966) and Copepods (Harding and Smith 1974).

Table 6. The density (no. $\mathrm{L}^{-1}$ ) of zooplankton taxa in five vertical net hauls in a random pattern from across Sand Lough on 19 June 2012. The haul length, arithmetic mean density, and CV and "composite" sample density calculated by weighting the relative volumes of water filtered. The lake area is 23.3 ha , maximum depth 7.2 m , mean depth 2.1 m , and $q$ value 2.4 .

| Haul | Length, m | Bosmina longirostris | Ceriodaphnia spp. | Cyclops spp. | Daphnia cuculatta | Daphnia galeata | Daphnia hyalina | Eudiaptmous gracilis |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 1 | 5.4 | 6.4 | 0.4 | 22.3 | 10.9 | 7.5 | 2.6 | 5.7 |
| 2 | 4.1 | 11.0 | 0.3 | 24.1 | 9.3 | 12.2 | 0.3 | 2.5 |
| 3 | 3.2 | 39.6 | 0 | 13.6 | 13.6 | 12.8 | 3.0 | 2.3 |
| 4 | 2.2 | 43.0 | 0 | 18.7 | 18.7 | 18.7 | 2.3 | 2.8 |
| 5 | 0.8 | 26.6 | 1.1 | 3.4 | 3.4 | 11.9 | 0 | 0.6 |
| Arithmetic mean |  | 25.32 | 0.36 | 16.42 | 11.18 | 12.62 | 1.64 | 2.78 |
| CV, \% |  | 65 | 125 | 51 | 50 | 32 | 85 | 66 |
| "Composite" |  | 11.20 | 0.25 | 4.40 | 3.47 | 5.14 | 0.42 | 0.71 |

Table 7. The density (no. $\mathrm{L}^{-1}$ ) of zooplankton taxa in five vertical net hauls in random pattern from across Lough Muckno on 22 May 2012. The haul length, arithmetic mean density, and CV and "composite" sample density calculated by weighting the relative volumes of water filtered. The lake area is 364 ha, maximum depth 27.0 m , mean depth 5.9 m , and $q$ value 3.6.

| Haul | Length, m | Bosmina coregoni | Bosmina longirostris | Chydorus spp. | Cyclops spp. | Daphnia hyalina | Daphnia hyalina var. galeata | Eudiaptmous gracilis |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 1 | 17.0 | 0.9 | 18.4 | 0 | 83.6 | 10.3 | 12.4 | 0 |
| 2 | 12.2 | 0 | 4.3 | 0.2 | 44.7 | 15.9 | 1.1 | 1.9 |
| 3 | 9.1 | 0 | 20.6 | 0 | 71.5 | 40.3 | 16.6 | 1.5 |
| 4 | 6.3 | 0 | 63.4 | 0 | 83.0 | 21.1 | 20.4 | 0.8 |
| 5 | 2.3 | 0.6 | 1.4 | 0 | 87.2 | 0 | 1.7 | 0.9 |
| Arithmetic mean |  | 0.30 | 21.62 | 0.04 | 74.00 | 17.52 | 10.44 | 1.02 |
| CV, \% |  | 141 | 115 |  | 24 | 85 | 84 | 71 |
| "Composite" |  | 0.12 | 6.52 | 0.01 | 30.86 | 4.40 | 3.00 | 0.37 |

## Assessment

The method was applied to four hypothetical lakes that represent the variation of lake form in the 51 lakes (Table 1) in order to establish its behavior. Each lake has an area of 10 ha and maximum depth 12 m , but the mean depths vary from 2.5 to 6.5 m and $q$ from 0.8 to 3.8 , so they cover a wide variation of lake form (Fig. 1).

The water depths of the five vertical net hauls in the four lakes are shown in Table 3. For a given $q$ value, varying the area does not change the sample depths.

How lake form influences the sample depths is shown in Fig. 4. As $q$ increases, the lake form becomes more convex (Fig. 1) and the water depths of the hauls decrease. This is a reflection of more of the lake volume being at shallower depths, as the hypsometric curve becomes more convex.

The proportions of the five hauls in the composite sample for the four lakes are shown in Table 4. While the method includes calculation of the relative contributions to the composite sample, there is limited variation over the four lakes, so
using the mean proportions (Table 4) would only introduce errors, based on the CV, of $8 \%$ to $32 \%$.

## Discussion

The method proposed here to select the water depths to sample zooplankton implemented a principle of sampling lake water that has been used by others, although we found their methods could only be applied in a general way (Blomqvist 2001) or there was insufficient detail provided for others to apply it (Girard et al. 2007; Rusak and Montz 2009). Therefore, our intention was to develop a well-defined method based on that principle and this was helped by using an empirical relationship for the hypsometric curve. While we used five samples to capture the variability of zooplankton abundance, as is typically employed, the method could be amended to use fewer or more samples. Based on the variability captured by five replicates, the method can be used in lakes up to a few tens of $\mathrm{km}^{2}$ is area.
Table 8. The density (no. $\mathrm{L}^{-1}$ ) of zooplankton taxa in five vertical net hauls in a random pattern from across Lower Lough MacNean on 15 May 2012. The haul length, arithmetic mean density, and CV and "composite" sample density calculated by weighting the relative volumes of water filtered. The lake area is 458 ha, maximum depth 12.1 m , mean depth 1.3 m , and $q$ value 8.3.


The selection of sampling sites is one part of characterizing the zooplankton in lakes. Frequency of sampling, including diurnal changes (Doubek et al. 2020), sampling equipment and sample enumeration also need to be considered. As has been noted, personal preferences, system constraints, and objectives of the investigation are usually involved (Mack et al. 2012), to which can be added the approach that is most common in the community within which the work is completed (Graham and Dayton 2002).

The zooplankton were sampled using vertical net hauls, but discrete samplers, tubes, pumps, etc., can also be employed (Bottrell et al. 1976; McCauley 1984; de Vries and Stein 1991). A 100\% filtration efficiency was assumed, but flow meters can be added (Bottrell et al. 1976; de Bernardi 1984). It could be that the shallowest sample occurs in the littoral zone of the lake and so a decision would have to be made about whether to use a vertical net haul or a more suitable method (Bottrell et al. 1976; de Vries and Stein 1991). A random pattern of samples at the depths indicated was used here, but a systematic one over the lake or another arrangement could also be used (Prepas 1984). While the swirling flask-Stempel pipette method is the most widely used subsample, the Folsom splitter and chambers are options (Sell and Evans 1982).

The method was used to estimate the whole-lake abundance (and biomass and mean length) of zooplankton in Spring, Summer, and Autumn in the 51 lakes (Table 1) and we encountered no practical difficulties applying it, such as Solver not producing a solution or unrealistic water depths of samples. The only issue that arose was the precision of taking a vertical net haul from a boat, as, occasionally, the haul length did not match exactly that indicated by the method; for example, around 2 m was used instead of the 2.4 m derived from the method. We also found, as noted, that the haul from the shallowest depth was occasionally in the littoral zone of a few lakes and we used a horizontal net tow of length equal to the water depth of Haul 5.

The method described here involves compositing the five samples in order to produce a single sample to represent the whole lake. However, each sample was analyzed separately in four of the lakes on one occasion and the detailed results are shown in Tables 5-8. The lakes cover a wide range of lake form: Legane Lough ( 6.0 ha ) is slightly concave and is a small lake; Sand (23.3 ha) and Muckno (364 ha) are convex, the typical form of most lakes, Sand a smaller and Muckno a larger lake; Lower Lough MacNean (458 ha) is very convex and a larger lake. All the lakes are eutrophic, with an annual mean Chl a concentration between 7.2 (Muckno) and 25.9 (Legane) $\mu \mathrm{g} \mathrm{L}^{-1}$.

The arithmetic mean taxon abundances in the five samples from each of the four lakes are generally greater than in the composite samples, the difference increasing as the density of zooplankton increases. This is due to compositing taking into account the different volumes of water filtered by the different haul lengths. This behavior may be general, as the variability
of abundance observed in the four lakes is similar to that found by others, although a little greater. In the four lakes, the coefficient of variation of the taxon abundance for composite sample abundance $\geq 1 \mathrm{~L}^{-1}$ varies from $24 \%$ to $85 \%$, with one value of $115 \%$, and a mean $(n=15)$ of $54.4 \%$ and median $50.5 \%$ (Tables 5-8). Based on a substantial collation of results from the literature, Downing et al. (1987) developed an empirical relationship between variance and mean density and their Eq. 2 gives a coefficient of variation between $38 \%$ and $54 \%$ for densities between 1 and $100 \mathrm{~L}^{-1}$.

## Comments and recommendations

The implementation in a spreadsheet of the method to select the water depths to take samples of zooplankton allows it to be applied with only information on the area, mean, and maximum depth of the lake needed. We found no practical problems using it to estimate the whole lake abundance of zooplankton in the 51 lakes, although, as there is no standard sampling method available, we could not assess its relative accuracy.

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[^0]:    *Correspondence: bhrt.rippey@ulster.ac.uk

